Legume Crops as Sources of Functional Compounds: Flavonoids in Seeds and Sprouts

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The recent development of the nutraceutical market is bringing up an increased demand of crops rich in compounds with beneficial effects on human health. Among these flavonoids, and more generally polyphenols, are an important class of plant secondary metabolites with proven bioactivity. The most appreciable sources of flavonoids in crops are grain legumes (particularly beans, chickpeas, peas), forage legumes (clover and alfalfa) and soybean. Numerous studies about flavonoid content have been performed on different genotypes of soybean, whereas available information about other legume species are quite scarce. In this context a scientific effort aimed at finding a "Mediterranean" alternative to soybean as source of functional compounds is of particular interest.
The objective of the present research was to screen a wide germplasm collection, composed of several accessions of grain legumes, for their phytochemical profiles in seeds and seedlings.

Methodology
35 grain legume accessions were investigated for their flavonoid content in both seeds and seedlings. For comparison 8 soybean (*Glycine max* L.) cultivars, known for their high flavonoid (isoflavones) content, were also included in the analysis The investigated plant material is part of a wider germplam collection composed of several accessions of forage and grain legumes available at the Department of Agroenvironmental Science and Technology, University of Bologna, Italy.
Flavonoids were extracted following the procedure described by Romani et al. (2005). Aglycone forms of glycoside flavonoids were obtained by acid hydrolysis of the extracts as previously described by Dinelli et al. (2006). The solutions were filtered through a 0.45 μm nylon filter prior to HPLC-DAD analyses.

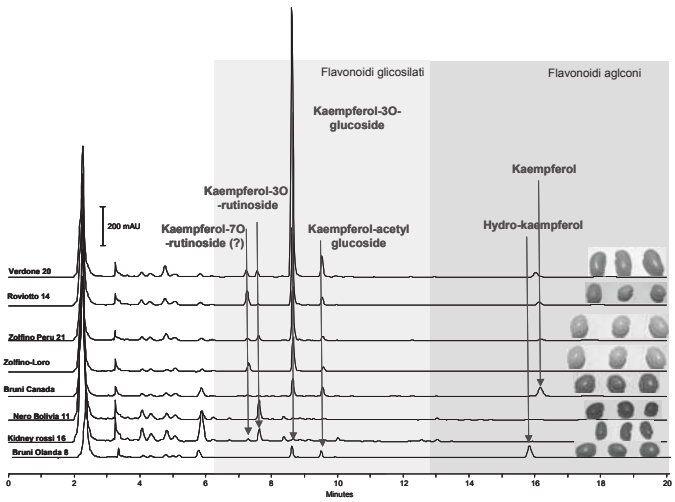

Figure 1: HPLC chromatograms of some of the investigated legume accessions highlighting peaks of glycosilated and aglycone flavonoids in seeds.

Results
Results highlighted a broad range of variability in bioactive compounds under both qualitative and quantitative composition. In particular, most of the seeds of investigated accessions contained conjugated forms of the flavonoid kaempferol (Figure 1). The compound kaempferol 3-O-glucoside was the main flavonoid found in all samples with a percentage ranging between 47% and 68% of total flavonoid content. The second most abundant flavonoid was kaempferol 3-O-xylosylglucoside with a percentage ranging between 16% and 33% of total flavonoid content.

Other bioactive compounds, such as vitexin, were also detected in seeds of adzuki bean accession (*Vigna angularis*). Beside one common bean accession (ie Zolfino del Perù) with a flavonoid content (1713.4 ± 11.6 µg/g seed weight) comparable with that detected in soybean seeds, the total flavonoid content of all other investigated accessions varied from 1.5 ± 0.5 (Bianco di Spagna) to 569.8 ± 2.6 (Roviotto) µg/g seed weight.

All accessions were also analyzed to identify and quantify flavonoids in sprouts. Daidzein, glycitein and genistein, along with kaempferol and quercetin were detected in most of analyzed accessions with amounts ranging from 2.8 to 86.7 µg/g fresh weight (Figure 2) (approximately 3-10 times lower than isoflavones in soybean sprouts).

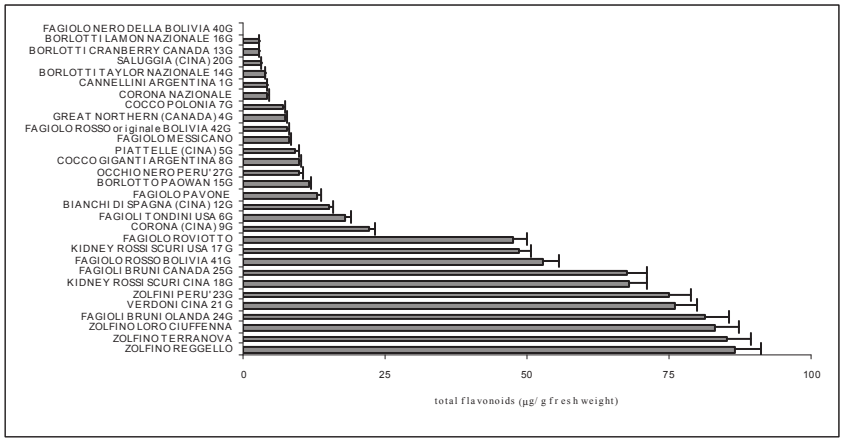

Figure 2. Total flavonoid content (µg/g fresh weight) detected in sprouts of some of the investigated legume accessions.

The great variability detected in the investigated accessions highlighted the marked incidence of the genotype in determining the functional compound content content of sprouts . The highest value were detected in the accessions Zolfino Reggello (86.79 ± 5.65 µg/g fresh weight), Zolfino Terranova Braccioloni (85.2 ± 4.6 µg/g fresh weight), Zolfino Loro Ciuffenna (83.1 ± 5.6 µg/g fresh weight), Bruno Olanda (81.4 ± 3.6 µg/g fresh weight) and Verdone Cina (75.9 ± 3.6 µg/g fresh weight). In these genotypes the flavonoid content is almost comparable to the total flavonoid content detected in soybean accessions (104.4 ± 5.8 µg/g fresh weight).

Conclusions

Despite many phytochemicals investigations on constituents of several leguminous species have been reported, studies focused mainly on the aerial parts (leaves, stems, flowers) of the plant (Williams et al., 1995). Data from the present study provided new insight on the flavonoid composition of several grain legume accessions in seeds. The identification of a bioactive compound, such as kaempferol, that may have health-promoting effects in humans, resulted to be of great interest in a perspective of source of functional compounds for nutraceutical application. In addition, to the best of our knowledge, this is the first time that sprouts of grain legumes other than soybean are proposed as sources of health-promoting phytochemicals (flavonoids).

To summarize, the present research highlighted the possibility of considering some of the investigated legume accessions as a valuable and "Mediterranean" alternative to soybean to obtain plant-derived products with health benefits.

References

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